Fabrication, Derivatization and Applications of Plastic Microfluidic Devices

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ABSTRACT

Control of the polymer surface chemistry is a crucial aspect in the development of plastic microfluidic devices. When commercially available plastic substrates are used to fabricate microchannels, differences in the electroosmotic flow (EOF) from plastic to plastic can be very high. Therefore, we have used polyelectrolyte multilayers (PEMs) to alter the surface of microchannels fabricated in plastics. The PEMs are easily fabricated and provide a means for controlling the flow direction and the electroosmotic mobility in the channels. Optimal modification of the microchannel surfaces was obtained by coating the channels with alternating layers of poly(allylamine hydrochloride) and poly(styrene sulfonate). The efficacy of the surface modification has been evaluated by measuring the electroosmotic flow mobility. When microchannels prepared in different polymer substrates were modified with PEMs, they demonstrated very similar electroosmotic mobilities. The PEMs have also been used to immobilize chemically selective molecules in the microchannels. In addition, relatively complex flow patterns, with simple arrangements of applied voltages, have been realized by derivatization of different arms of a single device with oppositely charged polyelectrolytes. Flow in opposite directions in the same channel is also possible; a positively derivatized plastic substrate with a negatively charged lid was used to achieve top-bottom opposite flows.

Keywords: Microfluidics, plastic, polyelectrolyte multilayers, electroosmotic flow, derivatization, imprinting

1. INTRODUCTION

Plastic microfluidic devices can be easily and inexpensively fabricated; however, the plastic surface chemical functionalities differ from those of glass and vary from polymer to polymer. Electrically driven flow is commonly used in microfluidics and the direction and rate of the electroosmotic flow (EOF) are a result of the substrate surface charge. Previous studies[1, 2] have shown that various polymer substrates support very different EOF mobilities in microchannel devices. In addition, the distribution of surface charge in imprinted plastic microchannels has been shown to be non-uniform.[3]

Modification of the chemical functionalities on the substrate surface can control surface charge and uniformity and offers methodology for controlling flow rate and direction in microfluidic devices. Numerous surface modification techniques have been developed for electrophoresis applications in silica and glass substrates. Glass and quartz microchannels have been derivatized previously using covalent[4, 5], non-covalent[6] and dynamic[7] coatings. In contrast, little research has focused on surface derivatization of plastics for microfluidic applications. Plastic microdevices have been derivatized using dynamic coating[8]; however, dynamic coatings lack long-term stability and require addition of the coating material to the running buffer.

We have recently shown that the deposition of polyelectroyte multilayers [9] (PEMs) is a simple, reproducible method for derivatization of plastic microfluidic devices.[10, 11] The multilayer is created by exposing the microchannel to alternating solutions of positively and negatively charged polyelectrolytes. Although the layers are adsorbed on the substrate or previous layer by noncovalent interactions, the resulting multilayers have multiple electrostatic bonds and are stable and uniform. PEMs were used to derivatize polystyrene (PS) and polyethylene terephthalate glycol (PETG) microchannels, resulting in comparable, reproducible EOF mobilities for the two substrates. Here we also demonstrate the utility of the coatings for incorporation of chemical selectivity and for control of the flow direction in microfluidic devices to achieve complex flow patterning and flow in opposite directions within a single channel.

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2. EXPERIMENTAL

2.1 Device preparation

Sheets of polystyrene, PS, (Corning Costar Corp., Cambridge, MA)[12], and poly(ethylene terephthalate glycol), PETG, (Vivak™, DMS Engineering Plastic Products, Sheffield, MA)[12], were cut into 7.6 cm by 7.6 cm squares and rinsed with methanol prior to use. Films of polydimethylsiloxane, PDMS, were made according to product information from a Sylgard 184™ silicone elastomer kit (Dow Corning, Midland, MI).[12] Figure 1 shows a schematic of the plastic microfluidic devices used in these studies. A silicon template, fabricated by photolithography[13], was used to imprint channels in the plastic substrates as previously described.[14] Each chip was imprinted with three cross or seven parallel channels, but only one is depicted in the figure for simplicity. Typical channels were 20 μm deep and 40 μm wide. All imprinted channels, including those modified with PEMs as described below, were sealed using a cured PDMS film. Holes 2 mm in diameter cut into the PDMS served as the fluid reservoirs for the channel. Two reservoirs, 2 to 4 cm apart were used in the straight channels, while four reservoirs, each 1 cm from the channel intersection were used in the cross design.

2.2 PEM deposition

An aqueous 60 mM poly(styrene sulfonate), sodium salt, PSS (Scientific Polymer Products, M_W = 500,000)[12] solution was prepared with 0.5 M NaCl and adjusted to pH 9 with sodium hydroxide (NaOH).[15] A 20 mM poly(allylamine hydrochloride), PAH (Aldrich, M_W = 70,000)[12] solution of the same salinity and pH[15] was also prepared. Polymer concentrations are based on the repeat unit. All water used in this study was deionized (18 MΩ•cm).

The PEM deposition method, similar to previously published methods of Chen and McCarthy[15], was used to deposit alternating layers of PAH and PSS. The substrate plastics, PS or PETG, were treated with 1M NaOH at 55°C for 15 min. The substrates were then rinsed with water and dried with nitrogen. The first PAH solution was pipetted onto the plastic substrate, completely covering the channels, and allowed to stand for 30 min. The PAH solution was removed by rinsing with water and dried with nitrogen. PSS was then pipetted on the plastic substrate, completely covering the channels, and allowed to stand for 30 min. The PSS was rinsed off with water and dried with nitrogen. Alternating layers of PAH and PSS were applied for 5 min with water rinses in between each solution application until the desired number of layers was deposited. Therefore, channels with an odd number of layers have a positively charged top layer, corresponding to PAH, while those with an even number of layers have a negative PSS surface.

2.3 Electroosmotic flow measurements

The EOF was measured using a current monitoring method[16] and the experimental details were published previously.[2] In the method, EOF is determined according to the equation, $\nu_{eof} = L*t^{-1}$, where L is the channel length and t is the time required for a second buffer of different concentration to fill the microchannel. The electroosmotic mobility is given by the ratio of the EOF rate to the applied field strength, E. The field strengths were typically 300 V/cm to 400 V/cm. Solutions containing 10 mM, 20 mM and 40 mM phosphate buffer, pH 7 were utilized for flow measurements in the 1) native plastic, 2) NaOH treated plastic, and 3) PEMs with the negative, PSS final layer. For measurements in channels with a final positive, PAH layer, 5 mM and 10 mM phosphate buffer solutions, pH 3, were used.

2.4 Fluorescence measurements

Fluorescent polystyrene beads, 1 μm in diameter, (Fluoresbrite™ [12]) were purchased from Polysciences, Inc.[12] Fluorescence imaging was performed using a research fluorescence microscope equipped with a mercury arc-lamp, appropriate filter sets, and a video camera (COHU)[12] for detection.

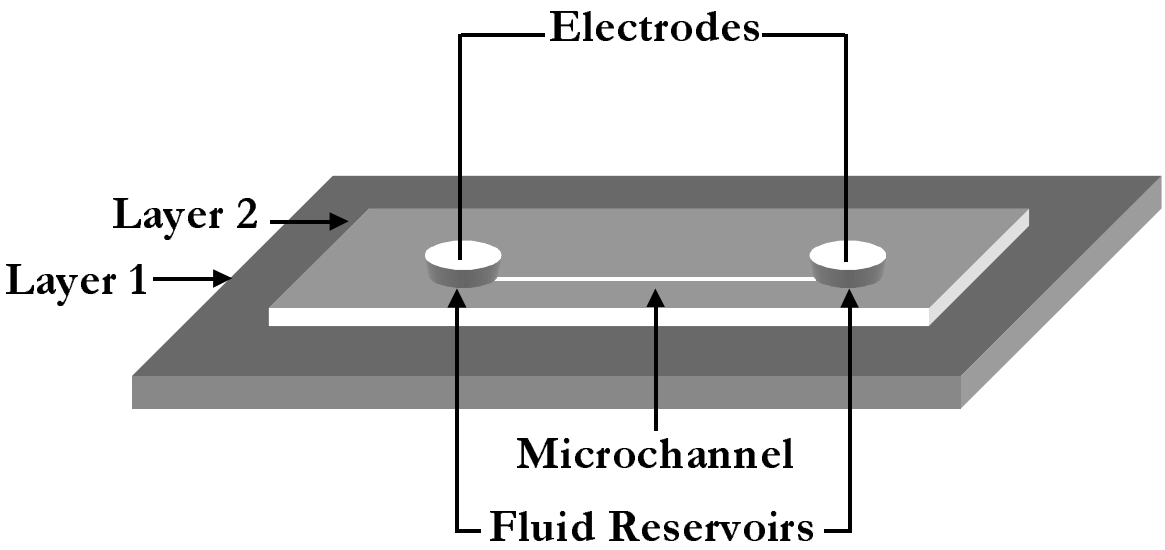

Figure 1. Plastic microfluidic device. Typical channel dimensions are 40 μm in width, 20 μm deep and 3 cm in length.

3. RESULTS AND DISCUSSION

While the use of plastics provides ease of fabrication and reduces costs, the plastic surface chemical functionalities differ from those of glass and vary from polymer to polymer. Differences in surface chemistry can have a dramatic effect on flow rates and separations in devices utilizing electroosmotic flow. Previous studies[1, 2] have shown that various polymer substrates support very different EOF mobilities in microchannel devices. In addition, the distribution of surface charge in imprinted plastic microchannels has been shown to be non-uniform.[3] Figure 2 shows channels imprinted by room temperature and hot imprinting and then exposed to a carboxylate-reactive dye. As can be see in the figure, the distribution of carboxylate groups is highly non-uniform in the room temperature imprinted channel and is quite different from the distribution seen in the hot imprinted channel.

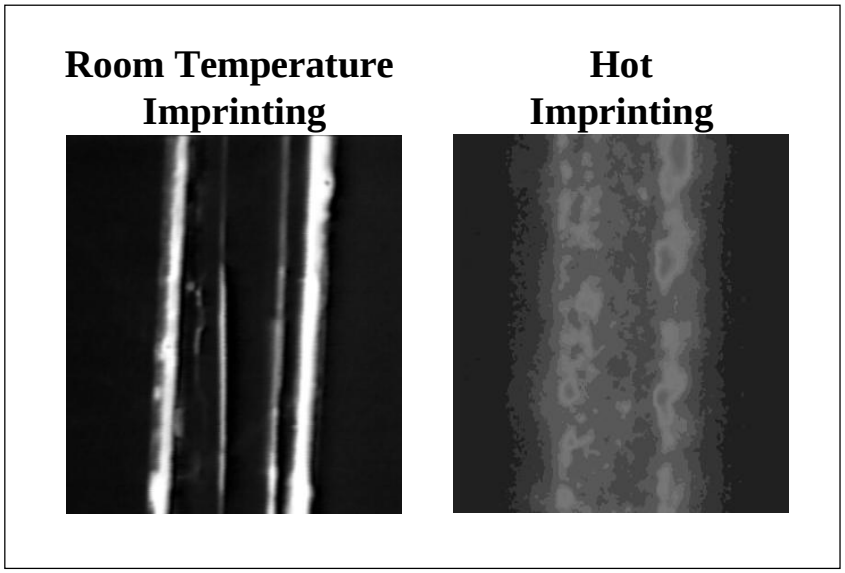

Figure 2. Fluorescence images of room temperature and hot imprinted microchannels labeled with carboxylate reactive fluorescent dye.

Polyelectroyte multilayers (PEMs) have been used to derivatize the surfaces of plastic microchannels to reduce surface charge variability in the channels. The flow direction and the electroosmotic flow mobility were used to characterize the PEM treated channels. The flow in channels with a negative PSS top layer was from anode to cathode, while the flow in channels with a positive PAH top layer was reversed and flowed from cathode to anode. The latter we designate as negative flow. The electroosmotic flow mobility was determined for a number of different layers, as shown in Figure 3. Standard deviations for repeated measurements on multiple channels were typically 10 % or less. For channels with a positive PAH top layer, the mobility was measured using pH 3 running buffer. For channels with the negative PSS top layer, the mobility was measured using pH 7 running buffer. The net EOF in channels with the negative PSS top layer was faster than, but in the opposite direction to, flow rates measured in channels with the positive PAH top layer. Video imaging, as described below, was used to determine that this slower net mobility was due to EOF in opposite directions within the channel. Graul and Schlenoff [18] reported slight drift in the electroosmotic flow mobility toward slower values for PEMs prepared in fused silica capillaries, if the capillaries were used continuously. Radiolabeling was used to determine that this drift resulted from conformational changes of the polyelectrolytes, rather than desorption from the substrate.[18] Slight mobility drift toward slower values was also observed in our work, but only with PEMs consisting of three layers on either substrate material. Such mobility drift was not observed for PEMs consisting of more than 3 layers. Deposition of 13 or 14 layers (Fig. 4) resulted in channels with similar mobilities in the PETG and PS, despite the differences in the mobility of the native plastics. This result indicates that the PEMs are a useful way of producing similar surfaces on different plastic materials, allowing greater flexibility in substrate polymer selection for a given application.

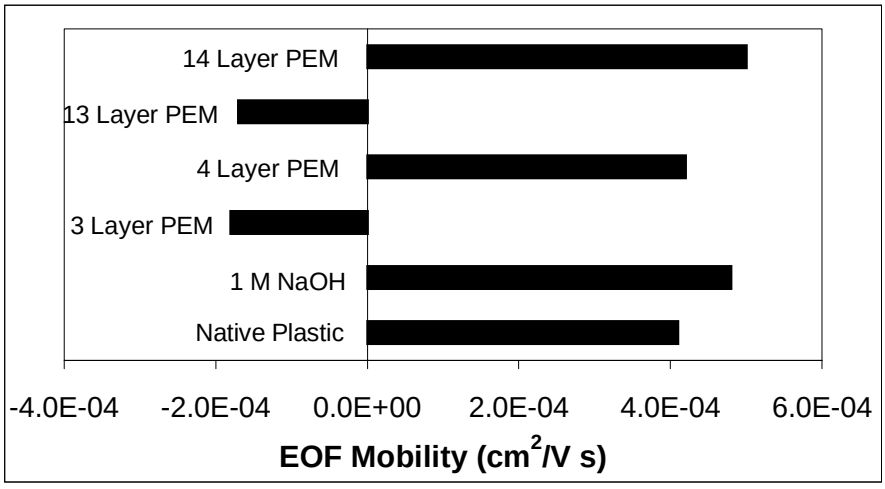

Figure 3. EOF mobility of PETG treated with PEMs of various numbers of layers.

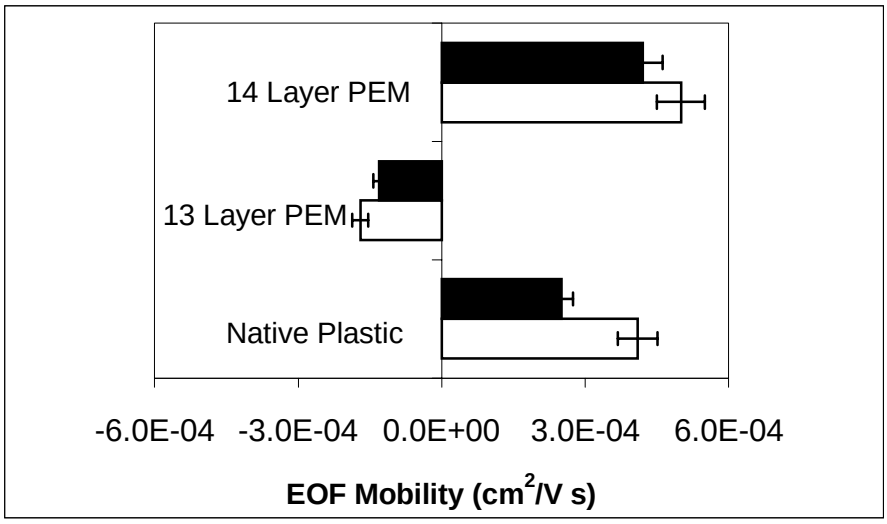

Figure 4. EOF mobility of untreated and PEMs treated PS and PETG. White bars: PETG; Black bars: PS.

The PEMs have also been used to incorporate chemical selectivity into the microchannels. PS channels were treated with a four layer PEM (PSS top layer). Streptavidin (with a net positive charge at neutral pH) was then allowed to adsorb to the negative PEM surface. The PEM treated streptavidin channels, PEM treated channels without streptavidin, and untreated, native plastic channels were exposed to fluorescently labeled biotin and then rinsed thoroughly. Figure 5 shows images of such channels. The native plastic channel appears black, as there was no measurable biotin in the channel. Slight non-specific adsorption of the biotin to the PEMs treated channel without the streptavidin is seen, but clearly the streptavidin-PEM had the highest affinity for the biotin. This indicates the efficacy of the PEMs for immobilizing sensing molecules.

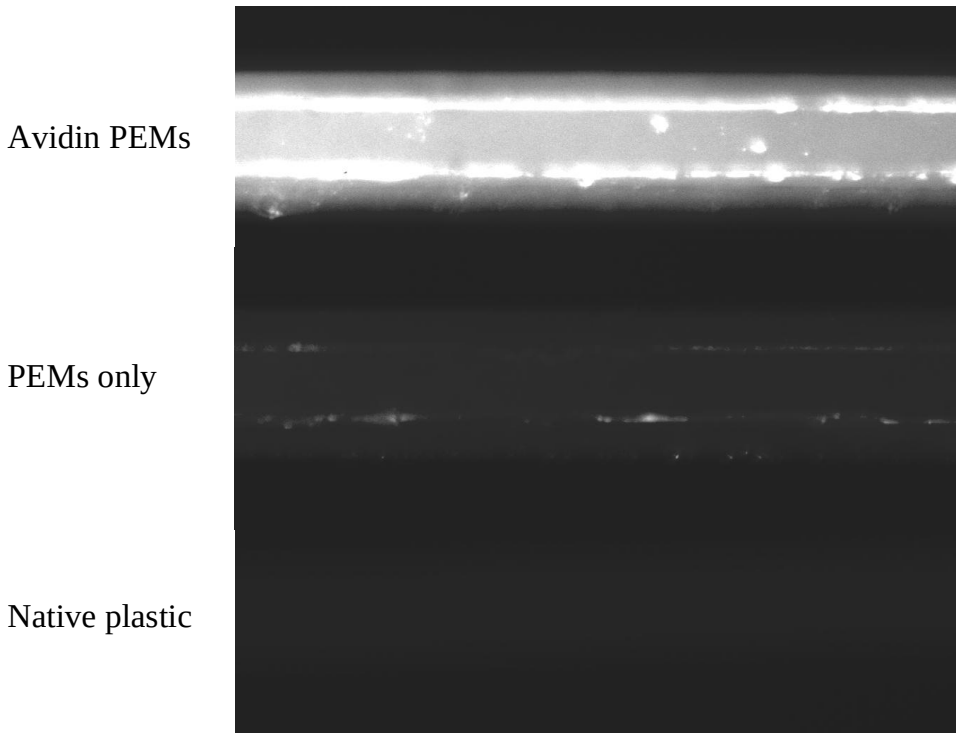

Figure 5. Fluorescence micrographs of 3 microchannels exposed to fluorescently-labeled biotin. The bottom, untreated plastic channel is not visible, as no labeled biotin is present in the channel. The middle channel was treated with PEMs, and small quantities of biotin non-specifically adsorbed to the PEM surface. The top channel exhibits the brightest fluorescence because it was treated with PEMs and streptavidin and, therefore, bound the highest level of biotin.

In microfluidic devices where electroosmotic flow is used, the flow direction depends on the wall charge. Since PEMs can be used to produce positively or negatively charged walls, they can be used to control the direction of the flow.[19] Figure 6 shows a schematic and a fluorescence image of flow in a cross channel in which three channels are negatively charged (PSS) and one channel is positively charged (PAH). Flow direction is depicted by the arrows in the schematic with a negative voltage applied on one channel and the other three channels grounded. Other patterns of derivatization, such as two positive arms and two negative arms have also been demonstrated, and the use of oppositely charged channels simplified the applied voltages needed for complicated flow patterns. In Fig. 6, a difference in flow velocity in the oppositely charged arms can also be seen. The flow in the three negatively charged arms appears faster, as evidenced by the blurred particle images, than in the positively charged arm. As described above, the EOF mobility in PS/PDMS derivatized with PEMs was determined and the net EOF in channels with the negative PSS top layer was faster than, but in the opposite direction to, flow rates measured in channels with the positive PAH top layer. Video imaging of the particles in the channels was used to determine that this slower net mobility was due to EOF in opposite directions within the PAH derivatized channels. The channel lids were not derivatized with the PEMs, and, therefore, the lids retained the negative surface charge of the native PDMS. These negative charges on the channel lid induce flow in the opposite direction of the flow propagated by the positive PAH layer on the polystyrene channel walls and bottom. The beads stopped when the applied voltage was switched off, indicating that flow was only electrically driven with no contribution from hydrodynamic flow.

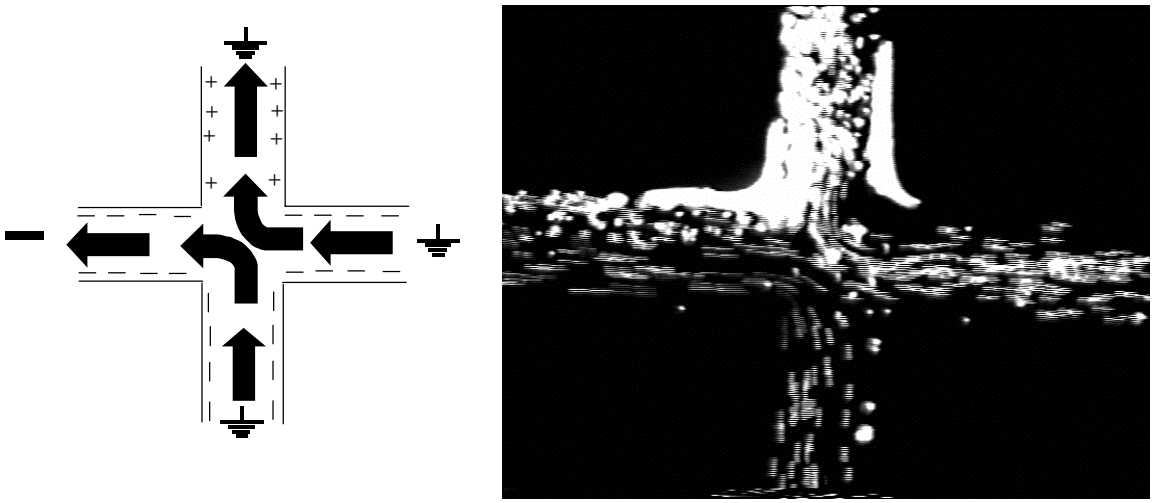

Figure 6. Schematic and fluorescence micrograph of bead movement in a cross device. Due to the beads' negative charge, bead adsorption to the channel wall is seen in the positively charged arm.

4. CONCLUSIONS

The PEMs were found to be an effective method for controlling the surface chemistry of plastic microchannels. Similar EOF mobilities were achieved in PETG and PS, despite the differences in the mobility of the native plastics. This result indicates that the PEMs are a useful way of producing similar surfaces on different plastic materials, allowing greater flexibility in substrate polymer selection for any given application. The PEMs can also be used to control the flow direction in microfluidic devices in order to achieve complex flow patterning and flow in opposite directions within a single channel. In addition, the utility of the PEMs as an immobilization matrix was demonstrated in the microchannels with a streptavidin-biotin complex. The avidin-biotin PEMs could potentially be used for the attachment of any biotinylated probe, and the protocol could be expanded to include incorporation of species other than avidin directly into the PEM.

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